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Recent Advances in Nicotinic Receptor Signaling in Alcohol Abuse and Alcoholism

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Abstract

Alcohol is the most commonly abused legal substance and alcoholism is a serious public health problem. It is a leading cause of preventable death in the world. The cellular and molecular mechanisms of alcohol reward and addiction are still not well understood. Emerging evidence indicates that unlike other drugs of abuse, such as nicotine, cocaine, or opioids, alcohol targets numerous channel proteins, receptor molecules, and signaling pathways in the brain. Previously, research has identified brain nicotinic acetylcholine receptors (nAChRs), a heterogeneous family of pentameric ligand-gated cation channels expressed in the mammalian brain, as critical molecular targets for alcohol abuse and dependence. Genetic variations encoding nAChR subunits have been shown to increase the vulnerability to develop alcohol dependence. Here, we review recent insights into the rewarding effects of alcohol, as they pertain to different nAChR subtypes, associated signaling molecules, and pathways that contribute to the molecular mechanisms of alcoholism and/or comorbid brain disorders. Understanding these cellular changes and molecular underpinnings may be useful for the advancement of brain nicotinic–cholinergic mechanisms, and will lead to a better translational and therapeutic outcome for alcoholism and/or comorbid conditions.

1. INTRODUCTION

Alcohol is the most commonly abused legal substance by humans. Alcoholism is a complex and chronic relapsing disorder that represents a serious global public health problem.¹ Alcoholism related deaths are estimated to account for 4% of all deaths worldwide and this number will be higher in the coming decades.² The prevalence of fetal alcohol syndrome, caused by mother's alcohol abuse and dependence, is also a significant problem in the United States and other industrialized nations.³ The estimated healthcare and economic cost from alcohol abuse and alcoholism in the United States, is significantly higher than many other diseases, including cancer.⁴ Thus, there is a need for better understanding of this complex brain disorder, for better therapeutic approaches to reduce alcohol abuse and relapse. Like nicotine addiction, emerging evidence suggests that neuronal nicotinic acetylcholine receptors (nAChRs), in the mesocorticolimbic-dopamine system, mediate the

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rewarding effects of alcohol and the development of alcohol dependence.^{5–9} Therefore, nAChRs are potential molecular targets for alcohol abuse and alcoholism. The primary focus of this review is to provide new insights into the rewarding effects of alcohol, as they pertain to different nAChR subtypes and other cholinergic factors that contribute to the molecular mechanisms of alcoholism. Understanding these cellular changes and molecular underpinnings, will lead to a better translational and therapeutic outcome of alcoholism. Considering the diversity of nAChRs in the brain, this chapter will discuss the role of different nAChR subunits, involving alcohol abuse and alcoholism obtained from genetic, preclinical, and clinical studies. In addition, the role of nAChRs in alcohol, nicotine coabuse, and other comorbid brain disorders will be discussed. The molecular targets and neurobiological mechanisms within the nAChRs system may offer a better understanding and effective therapeutic alternative to combat this complex relapsing brain disorder.

2. MESOLIMBIC-DOPAMINE SYSTEM AND nAChRs

Genetic, preclinical, and clinical studies have identified the brain nAChRs and membrane bound ion channels in the mesolimbic-dopamine system, as being involved in alcohol dependence.^{10–18} The nAChRs are pentameric ligand-gated cation channels with a diverse composition, which is widely expressed throughout the central nervous system (CNS).^{19–22} The nAChR-mediated signaling plays a critical role in many drug addiction processes, including the development of alcoholism.^{23–27} Twelve neuronal nAChR subunits, which are classified as either alpha subunits ($\alpha 2$ – $\alpha 10$) or beta subunits ($\beta 2$ – $\beta 4$), have been identified.^{19,28–31} These subunits assemble to form diverse functional nAChRs, which can be further subdivided into two major groups of brain nAChR subtypes.^{23,24,27} For example, heteromeric receptors are assembled from both alpha ($\alpha 2$ – $\alpha 6$) and beta subunits. The functional properties of such heteromeric receptors, depend on both the specific β and α subunits within the receptor complex and the β : α ratio of the subunits.^{28–31} Previous research indicates that the most abundant nAChRs in the CNS are $\beta 4\beta 2^*$, receptors containing both $\beta 4$ and $\beta 2$ subunits, and sometimes additional subunits (denoted by the asterisk). These receptors account for >90% of the high affinity nicotine binding sites in the brain.³⁰ The $\beta 4$ and $\beta 2$ subunits are colocalized in many parts of the brain, including thalamic nuclei, cortex, and ventral striatum.^{19,30}

On the other hand, homomeric receptors, such as, $\beta 7$ nAChR subtypes, are predominantly located in the hippocampal regions and cortical or subcortical regions, including the ventral striatum.³¹ In addition, neuronal nAChRs containing $\beta 6$ subunits are expressed in dopamine-rich areas (e. g., the mesocorticolimbic reward neurocircuit). Thus, $\beta 6^*$ receptors may be a new/novel drug target to treat many forms of drug addiction, including alcohol dependence.²⁵ A number of *invitro* or *invivo* studies have confirmed that alcohol activates the mesolimbic-dopamine system and elevates the synaptic release of dopamine in the ventral striatum, which partially mediates the rewarding effects of alcohol and other drugs of abuse.^{32–35} Therefore, mesocorticolimbic nAChRs are considered to be a molecular switch, activated by addictive behaviors.^{9,15,36–38} Neuronal nAChRs are widely expressed at the synapse, cell body, and axons in the CNS.²¹ Presynaptic nAChRs are involved in regulating the release of ACh, monoamines, and amino acids.^{39–43} In particular, dopamine release is

regulated by $\beta 4\beta 2\star$, $\beta 3\beta 2\star$, and $\beta 6\star$ nAChRs (\star indicates a possible involvement of other receptor subunits) in nigrostriatal/mesocorticolimbic terminals.^{44–47}

3. KNOCKOUT AND TRANSGENIC MODELS AND nAChRs

Recent reports with nAChR subtype knockout (KO) mice have provided important information about both brain nAChR function and the mediation of addiction-related behavior.^{23,48,49} For example, early research showed that mice lacking the $\beta 2$ subunit do not display too many nicotine-associated responses, including nicotine-induced DA release in the dorsal and ventral striatum, as well as nicotine-elicited increases in the firing rate of associated DA neurons.^{50,51} The lack of nicotine effect on the mesolimbic DA systems in $\beta 2$ subtype nAChR KO mice is consistent with the absence of nicotine self-administration by these animals.⁵¹ The $\beta 4$ subunit requires a $\beta 2$ subunit for assembly in a majority of heteromeric nAChRs in the brain. These and other studies using genetically modified mice suggest that $\beta 4/\beta 2\star$ nAChRs are critical for nicotine-related reward behaviors.^{52,53} Despite the distribution of the $\beta 7$ subunit in the brain, in particular its presence in the mesocorticolimbic system, studies in $\beta 7$ KO mice are not definitive about a role for the $\beta 7$ subunit in nicotine reward and conditioning.⁴⁸ However, $\beta 7\star$ nAChRs are important for long-term potentiation, neuroplasticity associated with learning, and memory in the mesolimbic reward pathway.⁴⁸ KO mouse studies targeting the $\beta 6$ subunit indicate that $\beta 6$ partners with $\beta 2$ nAChRs and may play an important role in nicotine addiction related behavior.⁵⁴ Like nicotine addiction, genetic studies have revealed that nAChRs are involved in ethanol self-administration, and reward behavior as well.¹¹ For example, a number of genetic studies have been conducted to identify the role of nAChR subtypes in ethanol-drinking behavior. Acute ethanol drinking behavior is reduced in $\beta 4$ KO mice, compared to wild type (WT), indicating a role for the nAChR $\beta 4\star$ subunit in ethanol abuse.^{11,26} Similarly, ethanol-related behavior and ethanol-induced midbrain dopaminergic function get decreased in $\beta 4$ KO mice.⁵⁵ On the other hand, $\beta 2$ KO mice behave similar to WT type mice in ethanol drinking behaviors.⁵⁶ In addition, $\beta 6$ KO and $\beta 3$ KO mice also display ethanol drinking behavior that is similar to WT mice in a two-bottle ethanol drinking paradigm.⁵⁶ Moreover, $\beta 7$ KO and WT mice consume similar amounts of ethanol, although there was a potential gender effect regarding $\beta 7$ nAChRs effects on ethanol consumption.⁵⁶ Again, $\beta 5$ KO mice do not differ in acute ethanol consumption, compared to WT mice.⁵⁷ Like nicotine-related behavior (as mentioned earlier), studies with transgenic over expression of the $\beta 5$, $\beta 3$, and $\beta 4$ receptor subunit genes indicate these subunits have a complex role in the modulation of ethanol-related behaviors.⁵⁸ Together, these data indicate that nAChRs containing $\beta 5$, $\beta 6$, $\beta 2$, or $\beta 3$ subunits may not be critical in ethanol drinking behaviors. Overall, the evidence suggests that $\beta 4$ receptors in the midbrain may be essential for ethanol-related behavior. These studies represent genetic mechanisms in ethanol dependence, involving brain nAChRs and associated neurobiological mechanisms.

It is widely recognized that brain nAChR subtypes are important mediators of the rewarding effects of ethanol and drugs of abuse.^{55,59–66} For example, systemic or local administration of nAChR ligands reduce ethanol drinking in a number of animal models.^{32,60,61,65} Furthermore, nAChRs in the ventral tegmental area (VTA) regulate ethanol consumption and associated mesolimbic neurochemical effects (e.g., dopamine release in the nucleus

accumbens (Acb)), as shown in various preclinical studies.^{60,62} However, some of these ligands produce mixed effects in alcohol/ethanol drinking behavior in humans,^{59,63} indicating mixed efficacy for treating ethanol dependence through nAChRs in the mesolimbic-dopamine system. Similarly, specific nAChR ligands may be resistant to pharmacologically efficacious reductions in ethanol drinking behavior, thus suggesting a role for other nAChR targets, such as $\beta 6\beta 2\star$.^{33,36} Additional studies with the $\beta 7$ nAChR ligand were found to be ineffective in reducing ethanol-taking behavior of animal models.⁵⁶ In other works, a partial $\beta 4\beta 2\star$ nAChR agonist was shown to reduce alcohol drinking in both animal models and humans.^{16,26,56,65,67–75} Given the mixed results with pharmacological efficacy on ethanol drinking behavior, the role of specific nAChR subtypes needs further investigation. Additional studies with cytisine, a partial agonist at $\beta 4\beta 2\star$ and lobeline, which is a nonselective antagonist, were found to reduce ethanol-taking behavior in a number of preclinical models.^{17,66,73–77} Interestingly, these nAChR ligands also altered alcohol-induced increases in mesolimbic tissue DA levels in mice,⁷⁷ supporting the important role of mesolimbic nAChRs in alcohol dependence. Confirming this, the nAChR ligands were found to reduce ethanol-taking behavior in a genetic animal model for alcohol abuse and dependence⁷⁸ or inbred mice,⁷⁴ indicating nAChRs are potential molecular targets for individuals with a genetic predisposition to develop alcohol dependence.

Thus, evidence suggests that selective desensitization of nAChRs with partial agonistic activity^{79,80} reduces ethanol-taking behavior in rats selectively bred for an alcohol preference.^{80,81} In addition with the involvement of nAChRs in ethanol-taking behavior, some of these ligands^{26,75,81,82} also decrease the alcohol deprivation effect, which is an animal model of relapse behavior.^{78,83–87} Emerging preclinical studies suggest that nicotine exposure reinstates alcohol seeking behaviors in rodents following extinction of alcohol reinforcement.^{88,89} Therefore, examination of cholinergic mechanisms associated with relapse is also important for new drug development to treat ethanol abuse and dependence.^{5,90} Overall, the existing animal and human studies suggest that ethanol-induced activation of the mesolimbic DA system involves brain diverse nAChRs stimulation, including $\beta 4\beta 2\star$, $\beta 6\beta 2\star$, as well as $\beta 3\beta 4\star$ subtypes.⁹¹ Thus, central nAChRs continue to be critical targets for the reinforcing and DA-activating effects of ethanol and underscore the need to conduct more subunit-specific nAChR research regarding ethanol abuse and dependence.

4. ALCOHOL, NICOTINE COABUSE, AND nAChRs

Due to the prevalence of ethanol and nicotine use,⁹² and the extremely high rates of smoking in individuals diagnosed with alcohol dependence,^{93,94,95} investigators have been working to develop models of coabuse in animals. However, till date very few such models have been validated. In one such model, within-session intravenous nicotine and oral ethanol drinking was demonstrated in Wistar rats.⁹⁶ More recently, ethanol-preferring (P) rats show oral operant intake of combined ethanol and nicotine solutions, that results in blood ethanol concentrations of approximately 100 mg% and blood nicotine levels of approximately 25 ng/mL. These levels are similar to levels obtained in human binge drinking and smoking.⁹⁷ Additionally, in adolescent C57BL/6 mice, exposure to cigarette smoke for 6 h/day for 16 days increased intake of 10% ethanol three to five fold in a scheduled access paradigm.⁹⁸

Also, chronic nicotine exposure in C3H mice during adolescence enhances ethanol withdrawal effects in adulthood.⁹⁹

In a study using rats selectively bred for high versus low locomotor activity induced by a novel field, it was found that adolescent exposure to nicotine facilitated an ethanol-induced conditioned place preference in early adulthood.¹⁰⁰ Regarding $\beta 4\beta 2\star$ receptors, a recent study revealed that chronic ethanol exposure/intake, including *in utero* exposure, by rhesus monkeys significantly decreased $\beta 4\beta 2\star$ levels in the frontal and insular cortex.¹⁰¹ In a study involving *in utero* exposure of rats, it was found that gestational exposure to ethanol and nicotine significantly increased nicotine self-administration during adolescence, and this effect seemed to be due to glutamatergic modulation of the mesolimbic-dopamine system.¹⁰² These data suggest a possible interaction between stages of development and cigarette smoke/nicotine exposure to increase ethanol and/or nicotine intake through different mechanisms that may extend into adulthood.

Animals selected for ethanol preference also show an increased sensitivity to the reinforcing effects of various drugs, including nicotine.⁹⁷ For instance, the P rat shows elevated sensitivity to the self-administration of intravenous nicotine, compared to the nonpreferring (NP) rat.¹⁰³ Consistent with this finding, P-rats are more sensitive than Wistar rats to the self-administration of nicotine directly into the posterior ventral tegmental area (pVTA) using the intracranial self-administration technique,¹⁰⁴ and ethanol and nicotine are co-self-administered into the pVTA at low concentrations that do not support self-administration individually.¹⁰⁵ However, in a recent study employing DBA/2J mice, nicotine was found to enhance the locomotor stimulating, but not the conditioned rewarding effects of ethanol, suggesting that in this model, combined stimulant effects of nicotine and ethanol do not predict enhanced reward.^{106,107}

Overall, current data suggest that ethanol and nicotine coabuse is rooted in the basic biological underpinnings of both drugs, supposedly by targeting $\beta 4\beta 2\star$ nAChR subtype in the mesolimbic-dopamine system. The drugs may function synergistically on some measures and each drug affects the other in terms of abuse liability. Emerging findings would suggest that the selection of ethanol-preference may increase the potential intake of nicotine coabuse, and adolescent coabuse may lead to elevated levels of coabuse in adulthood. Additional studies are clearly needed to provide a better understanding of the mechanisms that are involved in ethanol and nicotine coabuse.

5. GENETIC POLYMORPHISMS, ALCOHOL DEPENDENCE AND nAChRs

Emerging evidence indicates that cholinergic genes may play a significant role in ethanol-dependent behavior.¹⁰⁸ For example, significant associations between CHRNA6 polymorphisms (rs1072003, rs2304297, and rs892413) as well as CHRNB3 polymorphism (rs13280604) and excessive ethanol-drinking behavior have been reported.¹⁰⁹ In a recent study, using Hispanic and non-Hispanic white subjects from the Social and Emotional Contexts of Adolescent Smoking Patterns, revealed multiple polymorphisms of CHRNA4 were associated with a significantly elevated risk for adolescent binge drinking.¹¹⁰ These authors reported that polymorphisms for other nAChR genes were not associated with this

risk, which may be related to the ethnicity of the sample. Additional studies from the Nicotine Addiction Genetics consortium in Finland, reported a significant association between the CHRNA4 polymorphism rs11636753 and regular ethanol drinking with comorbidity for depression, which may have been sex dependent.¹¹¹ Regarding addiction in general, although polysubstance abuse may be a factor, the CHRNA5 risk polymorphism (rs16969968) is not only associated with nicotine dependence,¹¹² but also associated with other drugs of abuse, such as cocaine.¹¹³ Similarly, other CHRNA5 polymorphisms (rs615470 and rs684513) have significant associations with ethanol and cocaine dependence, respectively.¹¹³ Given an early onset of ethanol, drug use and abuse increases the probability of developing dependence later in life, it is important to examine whether these effects are under genetic control. One study examining polymorphisms within the CHRNA5-A3-B4 gene cluster found a significant association with the age of initiating the use of multiple abused substances.¹¹⁴ Similarly, recent findings from the San Diego Sibling Pair study indicate that, variants for CHRNA5 within this cluster are significantly associated with subjective level of response (i.e., intoxication) to ethanol.¹¹⁵ Interestingly, a recent study indicated missense variants in CHRNA3 may confer resistance to cocaine dependence in African Americans.¹¹⁶ However, it was also reported that a missense variant in CHRNA3 called rs149775276, was significantly associated with ethanol and cocaine dependence in European Americans.¹¹⁶ Combined findings indicate that multiple polymorphisms associated with nAChR genes and addictions, have been identified. Moreover, these associations predict dependence on a number of abused substances and/or associated behaviors, across national, ethnic, and psychiatric groups. Taken together, these studies suggest that nAChRs could be important targets for the development of therapeutics targeting multiple addictions, including alcoholism.

6. ALCOHOL DEPENDENCE, COMORBID PSYCHIATRIC CONDITION AND nAChRs

Recent evidence indicates that nAChRs are involved in drug addiction and comorbid psychiatric disorders, such as anxiety or depression.¹¹⁷ Just as in nicotine addiction,¹¹⁸ it has been proposed that there is a relationship between ethanol dependence and depression.^{119,120} For example, patients with depression have higher rates of ethanol-related problems, than the general population.^{121,122} Ethanol abstinence-related depression increases the chance of relapse because people may use ethanol for self-medication.¹²³ A number of studies have determined how genetic predisposition to high ethanol intake affects depression-like behavior, and how genetic predisposition to depression-like behavior affects ethanol intake in rats, but the results are not well established,¹²⁴ probably due to the challenges of defining criteria for valid animal models of these disorders. Previous studies suggest that chronic ethanol consumption in mice increases depression-like behavior during abstinence from ethanol.^{125,126} Therefore, alcoholism and depression supposedly share common molecular targets and associated neurobiological mechanisms. For example, abstinence from ethanol increases ACh release in the Acb,¹²⁷ and microdialysis studies indicate that ethanol abstinence induces rapid and sustained increases in extracellular ACh levels in the hippocampus.¹²⁸ A reduction in serotonergic neurotransmission has also been implicated in subpopulations of alcoholic patients.^{129,130} And, the efficacy of selective

serotonin reuptake inhibitors (SSRIs) provides strong support for the role of serotonin in depression, although SSRI treatment efficacy is highly variable for maintaining long-term abstinence from ethanol.¹³¹ Alcoholism,¹³² nicotine addiction,¹³³ and depression^{134,135} are disorders mediated by neuroplasticity including transcriptional control, especially with brain-derived neurotrophic factor (BDNF) and activity-regulated cytoskeleton-associated protein (Arc)¹³⁶ pathways. Regarding this, prolonged exposure to and abstinence from ethanol causes long-lasting neuroadaptations, which may underlie the development of depression-like behavior.¹²⁶ Moreover, chronic ethanol consumption reduces the expression of BDNF in the rat hippocampus, which is an effect seen in depression-like behavior in animal models.¹³⁷ Given the common neurobiological mechanisms in ethanol dependence and comorbid depression, it is possible that β_2 -nAChRs modulation and associated molecular mechanisms will be critical in reducing depression, in the presence of ethanol abuse and dependence. Therefore, the contribution of specific nAChR subtypes, and their molecular signaling in comorbid conditions, needs considerable additional investigation.

7. ALCOHOL, TRANSCRIPTION FACTORS, AND nAChRs

Evidence indicates that nAChRs modulate acute ethanol-induced increased expression of Fos family in the immediate early genes, such as c-Fos in midbrain dopaminergic neurons.^{17,26} In addition, both partial agonists and antagonists targeting nAChRs, reduce acute ethanol-induced c-Fos expression in the VTA and Acb.^{17,26} Similarly deltaFosB, which is a truncated splice variant of FosB gene, activity is associated with chronic ethanol exposure and neuroplasticity in the mesolimbic reward neurocircuit.^{138,139,140,141} Also, recent research suggests that nAChRs modulate deltaFosB upregulation in both ventral and dorsal striatum.⁷⁶ For example, partial agonists at nAChRs reduce chronic ethanol drinking behavior and its associated striatal deltaFosB upregulation.⁷⁶ Overall, these studies support the possible role of nAChRs in modulating long-lasting behavioral and molecular neuroadaptations that may be related to alcoholism. However, the role of nAChRs on other important transcription factors, such as cAMP-responsive element binding (CREB) expression requires further investigation.^{142,143}

8. CONCLUSIONS

In this review, evidence has been presented for the fact that AChRs in the mesolimbic-dopamine system are important molecular targets for ethanol abuse and alcoholism. It is evident from *in vitro* and *in vivo* studies that nAChRs are critically involved with synaptic activity of mesolimbic dopa-mine as well as associated cellular and molecular mechanisms, which underlie the addictive properties produced by ethanol. Given the variety of nAChR subtypes, localization and functions of these nAChRs are potential mediators of the complex neurobiological effects of ethanol. While the nAChR subtypes share a common structure, their pharmacological properties on drug addiction, including models of alcoholism, may depend on nAChR subunit composition. We believe that multiple nAChR subtypes could be targets for neurobiological effects of ethanol and their contribution to alcoholism. Therefore, understanding subtype-selective mechanisms will be critical for future translational research. Thus, brain nAChRs represent a potential molecular target for treating ethanol abuse and dependence. The fact that ethanol and nicotine addiction often cooccur in humans, is widely

recognized in this literature. Recent evidence indicates a genetic correlation between these two addictive disorders. Preclinical and clinical studies suggest that both nicotine and ethanol can, either directly or indirectly, activate the mesolimbic-dopamine system, which putatively mediates the rewarding effects and ethanol-induced addictive behavior, associated with nAChRs. Emerging data also indicate that neuronal nAChRs are involved in addiction and comorbid psychiatric disorders, such as anxiety or depression. Finally, it is important to note that nAChRs modulate important transcription factors, such as c-Fos or deltaFosB, in the mesolimbic-dopamine system that may be associated with chronic effects of ethanol. However, the role of nAChRs on CREB or BDNF expression associated with alcoholism remains to be determined. Overall, the evidence supporting a role for nAChRs in the neurobiological effects of ethanol is significant from a translational perspective, as are the implications of nAChRs in addictive behaviors, and a variety of comorbid psychiatric and cognitive conditions. Additional research and a refined understanding of the specific contribution of nAChR subtypes, and associated neurobiological mechanisms involving alcoholism and comorbid neuropsychiatric conditions, may identify new molecular targets and signaling pathways for the development of better treatment and prevention strategies.

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References

1. Dwyer-Lindgren L, Flaxman AD, Ng M, Hansen GM, Murray CJ, Mokdad AH. Drinking patterns in US counties from 2002 to 2012. *Am J Public Health*. 2015; 105:1120–1127. [PubMed: 25905846]
2. Report on Alcohol. Geneva: World Health Organization, Department of Mental Health and Substance Abuse; 2011.
3. Leibson T, Neuman G, Chudley AE, Koren G. The differential diagnosis of fetal alcohol spectrum disorder. *J Popul Ther Clin Pharmacol*. 2014; 21:e1–e30. [PubMed: 24639410]
4. CDC, Center for Disease Control. The Power of Prevention Chronic Disease. The Public Health Challenge of the 21st Century. Washington, DC: CDC; 2009.
5. Koob GF, Volkow ND. Neurocircuitry of addiction. *Neuropsychopharmacology*. 2010; 35:217–238. [PubMed: 19710631]
6. Volkow ND, Baler RD. Addiction science: uncovering neurobiological complexity. *Neuropharmacology*. 2014; 76(Pt B):235–249. [PubMed: 23688927]
7. Ericson M, Löf E, Stomberg R, Söderpalm B. The smoking cessation medication varenicline attenuates alcohol and nicotine interactions in the rat mesolimbic dopamine system. *J Pharmacol Exp Ther*. 2009; 329:225–230. [PubMed: 19126781]
8. Wise RA, Koob GF. The development and maintenance of drug addiction. *Neuropsychopharmacology*. 2014; 39:254–262. [PubMed: 24121188]
9. Rahman S, Engleman EA, Bell RL. Nicotinic receptor modulation to treat alcohol and drug dependence. *Front Neurosci*. 2015; 8:1–11.
10. Feduccia AA, Chatterjee S, Bartlett SE. Neuronal nicotinic acetylcholine receptor: neuroplastic changes underlying alcohol and nicotine addictions. *Front Mol Neurosci*. 2012; 5:1–18. [PubMed: 22319467]

11. Hendrickson LM, Guildford MJ, Tapper AR. Neuronal nicotinic acetylcholine receptors: common molecular substrates of nicotine and alcohol dependence. *Front Psychiatry*. 2013; 4:29. [PubMed: 23641218]
12. Rahman S, Prendergast MA. Brain cholinergic receptor system as a target for treating alcohol dependence. *Recent Patents CNS Drug Discov*. 2012; 7:145–150.
13. Rahman S. Nicotinic receptors as therapeutic targets for drug addictive disorders. *CNS Neurol Dis Drug Targets*. 2013; 12:633–640.
14. Wu J, Gao M, Taylor DH. Neuronal nicotinic acetylcholine receptors are important targets for alcohol reward and dependence. *Acta Pharmacol Sinica*. 2014; 35:311–315.
15. Chatterjee S, Bartlett SE. Neuronal nicotinic acetylcholine receptors as pharmacotherapeutic targets for the treatment of alcohol use disorders. *CNS Neurol Dis Drug Targets*. 2010; 9:60–76.
16. Chatterjee S, Steensland P, Rollema H, Bartlett SE, et al. Partial agonists of the alpha3 beta4 neuronal nicotinic acetylcholine receptor reduce ethanol consumption and seeking in rats. *Neuropsychopharmacology*. 2011; 36:603–615. [PubMed: 21048701]
17. Hendrickson LM, Zhao-Shea R, Tapper AR. Modulation of ethanol drinking-in-the-dark by mecamylamine and nicotinic acetylcholine receptor agonists in C57BL/6J mice. *Psychopharmacology*. 2009; 204:563–572. [PubMed: 19247637]
18. Tizabi Y, Copeland RL Jr, Louis VA, Taylor RE. Effects of combined systemic alcohol and central nicotine administration into ventral tegmental area on dopamine release in the nucleus accumbens. *Alcohol Clin Exp Res*. 2002; 26:394–399. [PubMed: 11923594]
19. Zoli M, Pistillo F, Gotti C. Diversity of native nicotinic receptor subtypes in mammalian brain. *Neuropharmacology*. 2015; 96:302–311. [PubMed: 25460185]
20. Millar NS, Gotti C. Diversity of vertebrate nicotinic acetylcholine receptors. *Neuropharmacology*. 2009; 56:237–246. [PubMed: 18723036]
21. Livingstone PD, Wonnacott S. Nicotinic acetylcholine receptors and the ascending dopamine pathways. *Biochem Pharmacol*. 2009; 78:744–755. [PubMed: 19523928]
22. Picciotto MR, Addy NA, Mineur YS, Brunzell DH. It is not “either/or”: activation and desensitization of nicotinic acetylcholine receptors both contribute to behaviors related to nicotine addiction and mood. *Prog Neurobiol*. 2008; 84:329–342. [PubMed: 18242816]
23. Fowler CD, Arends MA, Kenny PJ. Subtypes of nicotinic acetylcholine receptors in nicotine reward, dependence, and withdrawal: evidence from genetically modified mice. *Behav Pharmacol*. 2008; 19:461–484. [PubMed: 18690103]
24. Rahman S, Lopez-Hernandez G, Corrigan WA, Papke RL. Neuronal nicotinic receptors as brain targets for the pharmacotherapy of drug addiction. *CNS Neurol Dis Drug Targets*. 2008; 7:422–441.
25. Kuzmin A, Jerlhag E, Liljequist S, Engel JA. Effects of subunit selective nAChR receptors on operant ethanol self-administration and relapse-like ethanol-drinking behavior. *Psychopharmacology*. 2009; 203:99–108. [PubMed: 18987848]
26. Hendrickson LM, Zhao-Shea R, Pang X, Gardner PD, Tapper AR. Activation of alpha4* nAChRs is necessary and sufficient for varenicline-induced reduction of alcohol consumption. *J Neurosci*. 2010; 30:10169–10176. [PubMed: 20668200]
27. Tuesta LM, Fowler CD, Kenny PJ. Recent advances in understanding nicotinic receptor signaling mechanisms that regulate drug-self administration behavior. *Biochem Pharmacol*. 2011; 82:984–995. [PubMed: 21740894]
28. Le Novère N, Zoli M, Changeux JP. Neuronal nicotinic receptor alpha 6 subunit mRNA is selectively concentrated in catecholaminergic nuclei of the rat brain. *Eur J Neurosci*. 1996; 8:2428–2439. [PubMed: 8950106]
29. Buisson B, Bertrand D. Nicotine addiction: the possible role of functional upregulation. *Trends Pharmacol Sci*. 2002; 23:130–136. [PubMed: 11879680]
30. Lindstrom JM. Nicotinic acetylcholine receptors of muscles and nerves: comparison of their structures, functional roles, and vulnerability to pathology. *Ann NY Acad Sci*. 2003; 998:41–52. [PubMed: 14592862]
31. Gotti C, Clementi F. Neuronal nicotinic receptors: from structure to pathology. *Prog Neurobiol*. 2004; 74:363–396. [PubMed: 15649582]

32. Soderpalm B, Ericson M, Olausson P, Blomqvist O, Engel JA. Nicotinic mechanisms involved in the dopamine activating and reinforcing properties of ethanol. *Behav Brain Res*. 2000; 113:85–96. [PubMed: 10942035]
33. Larsson A, Svensson L, Söderpalm B, Engel JA. Role of different nicotinic acetylcholine receptors in mediating behavioral and neurochemical effects of ethanol in mice. *Alcohol*. 2002; 28:157–167. [PubMed: 12551757]
34. McBride WJ, Li TK. Animal models of alcoholism: neurobiology of high alcohol-drinking behavior in rodents. *Crit Rev Neurobiol*. 1998; 12:339–369. [PubMed: 10348615]
35. Tizabi Y, Bai L, Copeland RL Jr, Taylor RE. Combined effects of systemic alcohol and nicotine on dopamine release in the nucleus accumbens shell. *Alcohol*. 2007; 42:413–416.
36. Larsson A, Engel JA. Neurochemical and behavioral studies on ethanol and nicotine interactions. *Neurosci Biobehav Rev*. 2004; 27:713–720. [PubMed: 15019421]
37. Schlaepfer IR, Hoft NR, Ehringer MA. The genetic components of alcohol and nicotine co-addiction: from genes to behavior. *Curr Drug Abuse Rev*. 2008; 1:124–134. [PubMed: 19492010]
38. Crunelle CL, Miller ML, Booji J, Van den Brink W. The nicotinic acetylcholine receptor partial agonist varenicline and the treatment of drug dependence: a review. *Eur Neuropsychopharmacol*. 2010; 20:69–79. [PubMed: 19959340]
39. Wilkie GI, Hutson PH, Stephens MW, Whiting P, Wonnacott S. Hippocampal nicotinic autoreceptors modulate acetylcholine release. *Biochem Soc Trans*. 1993; 21:429–431. [PubMed: 8359505]
40. Alkondon M, Pereira EF, Barbosa CT, Albuquerque EX. Neuronal nicotinic acetylcholine receptor activation modulates γ -aminobutyric acid release from CA1 neurons of rat hippocampal slices. *J Pharmacol Exp Ther*. 1997; 283:1396–1411. [PubMed: 9400016]
41. Clarke P, Reuben M. Release of [3 H]-noradrenaline from rat hippocampal synaptosomes by nicotine: mediation by different nicotinic receptor subtypes from striatal [3 H]-dopamine release. *Br J Pharmacol*. 1996; 117:595–606. [PubMed: 8646402]
42. Grady S, Marks MJ, Wonnacott S, Collins AC. Characterization of nicotinic receptor-mediated [3 H] dopamine release from synaptosomes prepared from mouse striatum. *J Neurochem*. 1992; 59:848–856. [PubMed: 1494911]
43. Yang X, Criswell HE, Breese GR. Nicotine-induced inhibition in medial septum involves activation of presynaptic nicotinic cholinergic receptors on gamma-aminobutyric acid containing neurons. *J Pharmacol Exp Ther*. 1996; 276:482–489. [PubMed: 8632313]
44. Le Novère N, Zoli M, Changeux JP. Neuronal nicotinic receptor alpha 6 subunit mRNA is selectively concentrated in catecholaminergic nuclei of the rat brain. *Eur J Neurosci*. 1996; 8:2428–2439. [PubMed: 8950106]
45. Luo S, Kulak JM, Cartier GE, Jacobsen RB, Yoshikami D, Olivera BM, McIntosh JM. β -ConotoxinAulB selectively blocks $\beta 3\beta 4$ nicotinic acetylcholine receptors and nicotine-evoked norepinephrine release. *J Neurosci*. 1998; 18:8571–8579. [PubMed: 9786965]
46. Salminen O, Murphy KL, McIntosh JM, Drago J, Marks MJ, Collins AC, Grady SR. Subunit composition and pharmacology of two classes of striatal presynaptic nicotinic acetylcholine receptors mediating dopamine release in mice. *Mol Pharmacol*. 2004; 65:1526–1535. [PubMed: 15155845]
47. Wonnacott S, Kaiser S, Mogg A, Soliakov L, Jones IW. Presynaptic nicotinic receptors modulating dopamine release in the rat striatum. *Eur J Pharmacol*. 2000; 393:51–58. [PubMed: 10770997]
48. Mineur YS, Picciotto MR. Genetics of nicotinic acetylcholine receptors: relevance to nicotine addiction. *Biochem Pharmacol*. 2008; 75:323–333. [PubMed: 17632086]
49. Changeux JP. Nicotine addiction and nicotinic receptors: lessons from genetically modified mice. *Nat Rev Neurosci*. 2010; 11:389–401. [PubMed: 20485364]
50. Picciotto MR, Zoli M, Lena C, Bessis A, Lallemand Y, Le Novère N, et al. Abnormal avoidance learning in mice lacking functional high-affinity nicotine receptor in the brain. *Nature*. 1995; 374:65–67. [PubMed: 7870173]
51. Picciotto MR, Zoli M, Rimondini R, Lena C, Marubio LM, Pich EM, et al. Acetylcholine receptors containing the beta2 subunit are involved in the reinforcing properties of nicotine. *Nature*. 1998; 391:173–177. [PubMed: 9428762]

52. Ross SA, Wong JY, Clifford JJ, Kinsella A, Massalas JS, Horne MK, et al. Phenotypic characterization of an alpha 4 neuronal nicotinic acetylcholine receptor subunit knockout mouse. *J Neurosci.* 2000; 20:6431–6441. [PubMed: 10964949]
53. Tapper AR, McKinney SL, Nashmi R, Schwarz J, Deshpande P, Labarca C, et al. Nicotine activation of alpha4★ receptors: sufficient for reward, tolerance, and sensitization. *Science.* 2004; 306:1029–1032. [PubMed: 15528443]
54. Champiaux N, Gotti C, Cordero-Erausquin M, David DJ, Przybylski C, Léna C, Clementi F, Moretti M, Rossi FM, Le Novère N, McIntosh JM, Gardier AM, Changeux JP. Subunit composition of functional nicotinic receptors in dopaminergic neurons investigated with knock-out mice. *J Neurosci.* 2003; 23:7820–7829. [PubMed: 12944511]
55. Liu L, Hendrickson LM, Guildford MJ, Zhao-Shea R, Gardner PD, Tapper AR. Nicotinic acetylcholine receptors containing the $\beta 4$ subunit modulate alcohol reward. *Biol Psychiatry.* 2013; 73:738–746. [PubMed: 23141806]
56. Kamens HM, Anderson J, Picciotto MR. Modulation of ethanol consumption by genetic and pharmacological manipulation of nicotinic acetylcholine receptors in mice. *Psychopharmacology.* 2010; 208:613–626. [PubMed: 20072781]
57. Santos N, Chatterjee S, Henry A, Holgate J, Bartlett SE. The alpha5 neuronal nicotinic acetylcholine receptor subunit plays an important role in the sedative effects of ethanol but does not modulate consumption in mice. *Alcohol Clin Exp Res.* 2012; 37:655–662. [PubMed: 23164049]
58. Gallego X, Ruiz-Medina J, Valverde O, Molas S, Robles N, Sabrià J, Crabbe JC, Dierssen M. Transgenic over expression of nicotinic receptor alpha 5, alpha 3, and beta 4 subunit genes reduces ethanol intake in mice. *Alcohol.* 2012; 46:205–215. [PubMed: 22459873]
59. Blomqvist O, Ericson M, Johnson DH, Engel JA, Söderpalm B. Voluntary ethanol intake in the rat: effects of nicotinic acetylcholine receptor blockade or subchronic nicotine treatment. *Eur J Pharmacol.* 1996; 314:257–267. [PubMed: 8957244]
60. Ericson M, Blomqvist O, Engel JA, Söderpalm B. Voluntary ethanol intake in the rat and the associated accumbal dopamine overflow are blocked by ventral tegmental mecamylamine. *Eur J Pharmacol.* 1998; 358:189–196. [PubMed: 9822883]
61. Lê AD, Corrigan WA, Harding JW, Juzysch W, Li TK. Involvement of nicotinic receptors in alcohol self-administration. *Alcohol Clin Exp Res.* 2000; 24:155–163. [PubMed: 10698366]
62. Chi H, de Wit H. Mecamylamine attenuates the subjective stimulant-like effects of alcohol in social drinkers. *Alcohol Clin Exp Res.* 2003; 27:780–786. [PubMed: 12766622]
63. Young EM, Mahler S, Chi H, de Wit H. Mecamylamine and ethanol preference in healthy volunteers. *Alcohol Clin Exp Res.* 2005; 29:58–65. [PubMed: 15654292]
64. Reus VI, Obach RS, Coe JW, Faessel H, Rollema H, Watsky E, Reeves K. Varenicline: new treatment with efficacy in smoking cessation. *Drugs Today.* 2007; 43:65–75. [PubMed: 17353944]
65. Steensland P, Simms JA, Holgate J, Richards JK, Bartlett SE. Varenicline, an alpha4beta2 nicotinic acetylcholine receptor partial agonist, selectively decreases ethanol consumption and seeking. *Proc Natl Acad Sci USA.* 2007; 104:12518–12523. [PubMed: 17626178]
66. Bell RL, Eiler BJ, Cook JB, Rahman S. Nicotinic receptor ligands reduce ethanol intake by high alcohol-drinking HAD-2 rats. *Alcohol.* 2009; 43:581–592. [PubMed: 20004336]
67. McKee SA, Harrison EL, O'Malley SS, Krishnan-Sarin S, Shi J, Tetraault JM, et al. Varenicline reduces alcohol self-administration in heavy-drinking smokers. *Biol Psychiatry.* 2009; 66:185–190. [PubMed: 19249750]
68. Bito-Onon JJ, Simms JA, Chatterjee S, Holgate J, Bartlett SE. Varenicline, a partial agonist at neuronal nicotinic acetylcholine receptors, reduces nicotine-induced increases in 20% ethanol operant self-administration in Sprague-Dawley rats. *Addict Biol.* 2011; 16:440–449. [PubMed: 21392178]
69. Mitchell JM, Teague CH, Kayser AS, Bartlett SE, Fields HL. Varenicline decreases alcohol consumption in heavy-drinking smokers. *Psychopharmacology.* 2012; 223:299–306. [PubMed: 22547331]
70. Kaminski BJ, Weerts EM. The effects of varenicline on alcohol seeking and self-administration in baboons. *Alcohol Clin Exp Res.* 2014; 38:376–383. [PubMed: 24033702]

71. Litten RZ, Egli M, Heilig M, Cui C, Fertig JB, Ryan ML, Falk DE, Moss H, Huebner R, Noronha A. Medications development to treat alcohol dependence: a vision for the next decade. *Addict Biol.* 2012; 17:513–527. [PubMed: 22458728]
72. Sotomayor-Zarate R, Gysling K, Busto UE, Cassels BK, Tampier L, Quintanilla ME. Varenicline and cytosine: two nicotinic acetylcholine receptor ligands reduce ethanol intake in University of Chile bibulous rats. *Psychopharmacology.* 2013; 227:287–298. [PubMed: 23344555]
73. Sajja RK, Rahman S. Lobeline and cytosine reduce voluntary ethanol drinking behavior in male C57BL/6J mice. *Prog Neuropsychopharmacol Biol Psychiatry.* 2011; 35:257–264. [PubMed: 21111768]
74. Sajja RK, Rahman S. Neuronal nicotinic receptor ligands modulate chronic nicotine-induced ethanol consumption in C57BL/6J mice. *Pharmacol Biochem Behav.* 2012; 102:36–43. [PubMed: 22741175]
75. Sajja RK, Rahman S. Nicotinic receptor partial agonists modulate alcohol deprivation effect in C57BL/6J in mice. *Pharmacol Biochem Behav.* 2013; 110:161–167. [PubMed: 23872372]
76. Sajja RK, Rahman S. Cytosine modulates chronic voluntary ethanol consumption and ethanol-induced striatal up-regulation of Delta FosB in mice. *Alcohol.* 2013; 47:299–307. [PubMed: 23601929]
77. Sajja RK, Dwivedi C, Rahman S. Nicotinic ligands modulate ethanol-induced dopamine functions in mice. *Pharmacology.* 2010; 86:168–173. [PubMed: 20714207]
78. Bell RL, Sable HJK, Colombo G, Hyttia P, Rodd ZA, Lumeng L. Animal models for medications development targeting alcohol abuse using selectively bred rat lines: neurobiological and pharmacological validity. *Pharmacol Biochem Behav.* 2012; 103:119–155. [PubMed: 22841890]
79. Rezvani AH, Cauley M, Xiao Y, Kellar KJ, Levin ED. Effects of chronic sazetidine-A, a selective $\beta 4 \beta 2$ neuronal nicotinic acetylcholine receptors desensitizing agent on pharmacologically-induced impaired attention in rats. *Psychopharmacology.* 2013; 226:35–43. [PubMed: 23100170]
80. Xiao Y, Fan H, Musachio JL, Wei ZL, Chellappan SK, Kozikowski AP, et al. Sazetidine-A, a novel ligand that desensitizes $\beta 4 \beta 2$ nicotinic acetylcholine receptors without activating them. *Mol Pharmacol.* 2006; 70:1454–1460. [PubMed: 16857741]
81. Rezvani AH, Slade S, Wells C, Petro A, Lumeng L, Li T-K, et al. Effects of sazetidine-A a selective $\alpha 4 \beta 2$ nicotinic acetylcholine receptor desensitizing agent on alcohol and nicotine self-administration in selectively bred alcohol-preferring (P) rats. *Psychopharmacology.* 2010; 211:161–174. [PubMed: 20535453]
82. Wouda JA, Riga D, De Vries W, Stegeman M, van Mourik Y, Schetters D, et al. Varenicline attenuates cue-induced relapse to alcohol but not nicotine seeking while reducing inhibitory response control. *Psychopharmacology.* 2011; 216:267–277. [PubMed: 21331520]
83. McKinzie DL, Nowak KL, Yorger L, McBride WJ, Murphy JM, Lumeng L, et al. The alcohol deprivation effect in the alcohol preferring P rat under free-drinking and operant conditions. *Alcohol Clin Exp Res.* 1998; 22:1170–1176. [PubMed: 9726292]
84. Rodd ZA, Bell RL, Sable HJK, Murphy JM, McBride WJ. Recent advances in animal models of alcohol craving and relapse. *Pharmacol Biochem Behav.* 2004; 79:439–450. [PubMed: 15582015]
85. Spanagel R, Höltner SM. Long-term alcohol self-administration with repeated alcohol deprivation phases: an animal model of alcoholism? *Alcohol.* 1999; 34:231–243.
86. Sparta DR, Ferraro FM III, Fee JR, Knapp DJ, Breese GR, Thiele TE. The alcohol deprivation effect in C57BL/6J mice is observed using operant self-administration procedures and is modulated by CRF-1 receptor signaling. *Alcohol Clin Exp Res.* 2009; 33:31–42. [PubMed: 18945225]
87. Melendez RI, Middaugh LD, Kalivas PW. Development of an alcohol deprivation and escalation effect in C57BL/6J mice. *Alcohol Clin Exp Res.* 2006; 30:2017–2025. [PubMed: 17117967]
88. Lê AD, Li Z, Funk D, Shram M, Li T-K, Shaham Y. Nicotine increases alcohol self-administration and reinstates alcohol seeking in rats. *Psychopharmacology.* 2003; 68:216–221. [PubMed: 12536264]
89. Hauser SR, Getachew B, Oster SM, Dhaher R, Ding Z-M, Bell RL, et al. Nicotine modulates alcohol-seeking and relapse by alcohol-preferring (P) rats in a time dependent manner. *Alcohol Clin Exp Res.* 2012; 36:43–54. [PubMed: 21689122]

90. Weiss F, Porrino LJ. Behavioral neurobiology of alcohol addiction: recent advances and challenges. *J Neurosci*. 2002; 22:3332–3337. [PubMed: 11978808]
91. Rollema H, Coe JW, Chambers LK, Hurst RS, Stahl SM, Williams KE. Rationale, pharmacology and clinical efficacy of partial agonists of alpha4beta2 nACh receptors for smoking cessation. *Trends Pharmacol Sci*. 2007; 28:316–325. [PubMed: 17573127]
92. Kandel D, Chen K, Warner LA, Kessler RC, Grant B. Prevalence and demographic correlates of symptoms of last year dependence on alcohol, nicotine, marijuana and cocaine in the U.S. population. *Drug Alcohol Depen*. 1997; 44:11–29.
93. Gulliver SB, Rohsenow DJ, Colby SM, Dey AN, Abrams DB, Niaura RS, et al. Interrelationship of smoking and alcohol dependence, use and urges to use. *J Stud Alcohol*. 1995; 56:202–206. [PubMed: 7760567]
94. Hughes JR. Treating smokers with current or past alcohol dependence. *Am J Health Behav*. 1996; 20:286–290.
95. John U, Meyer C, Rumpf H, Schumann JA, Thyrian JR, Hapke U. Strength of the relationship between tobacco smoking, nicotine dependence and the severity of alcohol dependence syndrome criteria in a population-based sample. *Alcohol Alcohol*. 2003; 38:606–612. [PubMed: 14633650]
96. Lê AD, Lo S, Harding S, Juzysch W, Marinelli PW, Funk D. Co-administration of intravenous nicotine and oral alcohol in rats. *Psychopharmacology*. 2010; 208:475–486. [PubMed: 20013113]
97. McBride WJ, Rodd ZA, Bell RL, Lumeng L, Li TK. The alcohol-preferring (P) and high-alcohol-drinking (HAD) rats—animal models of alcoholism. *Alcohol*. 2014; 48:209–215. [PubMed: 24268381]
98. Burns BE, Proctor WR. Cigarette smoke exposure greatly increases alcohol consumption in adolescent C57BL/6 mice. *Alcohol Clin Exp Res*. 2013; 37:E364–E372. [PubMed: 22827559]
99. Riley HH, Zalud AW, Diaz-Granados JL. The influence of a chronic adolescent nicotine exposure on ethanol withdrawal severity during adulthood in C3H mice. *Alcohol*. 2010; 44:81–87. [PubMed: 20113876]
100. Philpot RM, Engberg ME, Wecker L. Ethanol conditioned place preference and alterations in deltaFosB following adolescent nicotine administration differ in rats exhibiting high or low behavioral reactivity to a novel environment. *Behav Brain Res*. 2014; 262:101–108. [PubMed: 24412683]
101. Hillmer AT, Tudorascu DL, Wooten DW, Lao PJ, Barnhart TE, Ahlers EO, Resch LM, Larson JA, Converse AK, Moore CF, Schneider ML, Christian BT. Changes in the alpha4beta2★ nicotinic acetylcholine system during chronic controlled alcohol exposure in nonhuman primates. *Drug Alcohol Depen*. 2014; 138:216–219.
102. Roguski EE, Sharp BM, Chen H, Matta SG. Full-gestational exposure to nicotine and ethanol augments nicotine self-administration by altering ventral tegmental dopami-nergic function due to NMDA receptors in adolescent rats. *J Neurochem*. 2014; 128:701–712. [PubMed: 24147868]
103. Lê AD, Li Z, Funk D, Shram M, Li TK, Shaham Y. Increased vulnerability to nicotine self-administration and relapse in alcohol-naïve offspring of rats selectively bred for high alcohol intake. *The J Neurosci*. 2006; 26:1872–1879. [PubMed: 16467536]
104. Hauser SR, Bracken AL, Deehan GA Jr, Toalston JE, Ding ZM, Truitt WA, Bell RL, McBride WJ, Rodd ZA. Selective breeding for high alcohol preference increases the sensitivity of the posterior VTA to the reinforcing effects of nicotine. *Addict Biol*. 2014; 19:800–811. [PubMed: 23496648]
105. Truitt WA, Hauser SR, Deehan GA Jr, Toalston JE, Wilden JA, Bell RL, McBride WJ, Rodd ZA. Ethanol and nicotine interaction within the posterior ventral tegmental area in male and female alcohol-preferring rats: evidence of synergy and differential gene activation in the nucleus accumbens shell. *Psychopharmacology*. 2015; 232:639–649. [PubMed: 25155311]
106. Gubner NR, McKinnon CS, Reed C, Phillips TJ. Accentuating effects of nicotine on ethanol response in mice with high genetic predisposition to ethanol-induced locomotor stimulation. *Drug Alcohol Depend*. 2013; 127:108–114. [PubMed: 22795175]
107. Gubner NR, Cunningham CL, Phillips TJ. Nicotine enhances the locomotor stimulating but not the conditioned rewarding effect of ethanol in DBA/2J mice. *Alcohol Clin Exp Res*. 2015; 39:64–72. [PubMed: 25623407]

108. Madden PA, Heath AC. Shared genetic vulnerability in alcohol and cigarette use and dependence. *Alcohol Clin Exp Res.* 2002; 26:1919–1921. [PubMed: 12500120]
109. Hoft NR, Corley RP, McQueen MB, Huizinga D, Menard S, Ehringer MA. SNPs in CHRNA6 and CHRNA3 are associated with alcohol consumption in a nationally representative sample. *Genes Brain Behav.* 2009; 8:631–637. [PubMed: 19500157]
110. Coon H, Piasecki TM, Cook EH, Dunn D, Mermelstein RJ, Weiss RB, Cannon DS. Association of the CHRNA4 neuronal nicotinic receptor subunit gene with frequency of binge drinking in young adults. *Alcohol Clin Exp Res.* 2014; 38:930–937. [PubMed: 24428733]
111. Broms U, Wedenoja J, Largeau MR, Korhonen T, Pitkaniemi J, Keskitalo-Vuokko K, Hoppola A, Heikkilä KH, Heikkilä K, Ripatti S, Sarin AP, et al. Analysis of detailed phenotype profiles reveals CHRNA5-CHRNA3-CHRNA4 gene cluster association with several nicotine dependence traits. *Nic Tob Res.* 2012; 4:720–733.
112. Anantharaman D, Chabrier A, Gaborieau V, Franceschi S, Herrero R, Rajkumar T, Samant T, Mahimkar MB, Brennan P, McKay JD. Genetic variants in nicotine addiction and alcohol metabolism genes, oral cancer risk and the propensity to smoke and drink alcohol: a replication study in India. *PLoS ONE.* 2014; 9:e88240.
113. Sherva R, Kranzler HR, Yu Y, Logue MW, Poling J, Arias AJ, Anton RF, Oslin D, Farrer LA, Gelernter J. Variation in nicotinic acetylcholine receptor genes is associated with multiple substance dependence phenotypes. *Neuropsychopharmacology.* 2010; 35:1921–1931. [PubMed: 20485328]
114. Lubke GH, Stephens SH, Lessem JM, Hewitt JK, Ehringer MA. The CHRNA5/A3/B4 gene cluster and tobacco, alcohol, cannabis, inhalants and other substance initiation: replication and new findings using mixture analyses. *Behav Genet.* 2012; 42:636–646. [PubMed: 22382757]
115. Choquet H, Joslyn G, Lee A, Kasberger J, Robertson M, Brush G, Schuckit MA, White R, Jorgenson E. Examination of rare missense variants in the CHRNA5-A3-B4 gene cluster to level of response to alcohol in the San Diego Sibling Pair study. *Alcohol Clin Exp Res.* 2013; 37:1311–1316. [PubMed: 23458267]
116. Haller G, Kapoor M, Budde J, Xuei X, Edenberg H, Nurnberger J, Kramer J, Brokks A, Tischfield J, Almasy L, Agrawal A, Bucholz K, Rice J, Saccone N, Bierut L, Goate A. Rare missense variants in CHRNA3 and CHRNA5 are associated with risk of alcohol and cocaine dependence. *Hum Mol Genet.* 2014; 23:810–819. [PubMed: 24057674]
117. Rahman S. Targeting brain nicotinic acetylcholine receptors to treat major depression and comorbid alcohol or nicotine addiction. *CNS Neurol Disord Drug Targets.* 2015; 14:647–653. [PubMed: 25921743]
118. Roni MA, Rahman S. The effects of lobeline on nicotine withdrawal-induced depression-like behavior in mice. *Psychopharmacology.* 2014; 231:2989–2998. [PubMed: 24682499]
119. Sullivan LE, Fiellin DA, O'Connor PG. The prevalence and impact of alcohol problems in major depression: a systematic review. *Am J Med.* 2005; 118:330–341. [PubMed: 15808128]
120. Boschloo L, Vogelzangs N, Smit JH, van den Brink W, Veltman DJ, Beekman ATF, Penninx BWJH. Comorbidity and risk indicators for alcohol use disorders among persons with anxiety and/or depressive disorders: findings from the Netherlands Study of Depression and Anxiety (NESDA). *J Affect Disord.* 2011; 131:233–242. [PubMed: 21247636]
121. Kessler RC, Nelson CB, McGonagle KA, Edlund MJ, Frank RG, Leaf PJ. The epidemiology of co-occurring addictive and mental disorders: implications for prevention and service utilization. *Am J Orthopsychiatry.* 1996; 66:17–31. [PubMed: 8720638]
122. Garbutt JC, West SL, Carey TS, Lohr KN, Crews FT. Pharmacological treatment of alcohol dependence. *JAMA.* 1999; 281:1318–1325. [PubMed: 10208148]
123. Hasin DS, Grant BF. Major depression in 6050 former drinkers: association with past alcohol dependence. *Arch Gen Psychiatry.* 2002; 59:794–800. [PubMed: 12215078]
124. Godfrey CD, Froehlich JC, Stewart RB, Li TK, Murphy JM. Comparison of rats selectively bred for high and low ethanol intake in a forced-swim-test model of depression: effects of desipramine. *Physiol Behav.* 1997; 62:729–733. [PubMed: 9284491]

125. Pang TY, Renoir T, Du X, Lawrence AJ, Hannan AJ. Depression-related behaviours displayed by female C57BL/6J mice during abstinence from chronic ethanol consumption are rescued by wheel running. *Eur J Neurosci.* 2013; 37:1803–1810. [PubMed: 23551162]
126. Stevenson JR, Schroeder JP, Nixon K, Besheer J, Crews FT, Hodge CW. Abstinence following alcohol drinking produces depression-like behavior and reduced hippocampal neurogenesis in mice. *Neuropsychopharmacology.* 2009; 34:1209–1222. [PubMed: 18563059]
127. Rada P, Johnson D, Lewis M, Hoebel B. In alcohol-treated rats, naloxone decreases extracellular dopamine and increases acetylcholine in the nucleus accumbens: evidence of opioid withdrawal. *Pharmacol Biochem Behav.* 2004; 79:599–605. [PubMed: 15582668]
128. Imperato A, Dazzi L, Carta G, Colombo G, Biggio G. Rapid increase in basal acetylcholine release in the hippocampus of freely moving rats induced by withdrawal from long-term ethanol intoxication. *Brain Res.* 1998; 784:347–350. [PubMed: 9518686]
129. Patkar AA, Gopalakrishnan R, Naik PC, Murray HW, Vergare MJ, Marsden CA. Changes in plasma noradrenaline and serotonin levels and craving during alcohol withdrawal. *Alcohol.* 2003; 38:224–231.
130. Cadete-Leite A, Brandao F, Andrade J, Ribeiro-da-Silva A, Paula-Barbosa M. The GABAergic system of the dentate gyrus after withdrawal from chronic alcohol consumption: effects of intracerebral grafting and putative neuroprotective agents. *Alcohol.* 1997; 32:471–484.
131. Narnajo CA, Knoke DM. The role of selective serotonin reuptake inhibitors in reducing alcohol consumption. *J Clin Psychiatry.* 2001; 62:18–25.
132. Yakovleva T, Bazov I, Watanabe H, Hauser KF, Bakalkin G. Transcriptional control of maladaptive and protective responses in alcoholics: a role of the NF- κ B system. *Brain Behav Immun.* 2011; 25:S29–S38. [PubMed: 21195164]
133. Gao Y, Peng S, Wen Q, Zheng C, Lin J, Tan Y, Ma Y, Luo Y, Xue Y, Wu P, Ding Z, Lu L, Li Y. The mammalian target of rapamycin pathway in the basolateral amygdala is critical for nicotine-induced behavioural sensitization. *Intl J Neuropsychopharmacol.* 2014; 17:1881–1894.
134. Carlberg L, Scheibelreiter J, Hassler MR, Schloegelhofer M, Schmoeger M, Ludwig Kasper S, Aschauer H, Egger G, Schosser A. Brain-derived neurotrophic factor (BDNF)—epigenetic regulation in unipolar and bipolar affective disorder. *J Affect Dis.* 2014; 168:399–406. [PubMed: 25106037]
135. Mitchelmore C, Gede L. Brain derived neurotrophic factor: epigenetic regulation in psychiatric disorders. *Brain Res.* 2014; 1586:162–172. [PubMed: 25223903]
136. Rosi S. Neuroinflammation and the plasticity-related immediate-early gene. *Arc Brain Behav Immun.* 2011; 25:S39–S49. [PubMed: 21320587]
137. Hauser SR, Getachew B, Taylor RE, Tizabi Y. Alcohol induced depressive-like behavior is associated with a reduction in hippocampal BDNF. *Pharmacol Biochem Behav.* 2011; 100:253–258. [PubMed: 21930150]
138. McClung CA, Nestler EJ. Neuroplasticity mediated by altered gene expression. *Neuropsychopharmacol Rev.* 2008; 33:3–17.
139. Li J, Cheng Y, Bian W, Liu X, Zhang C, Ye JH. Region-specific induction of FosB/ FosB by voluntary alcohol intake: effects of naltrexone. *Alcohol Clin Exp Res.* 2010; 34:1742–1750. [PubMed: 20626732]
140. Robison AJ, Nestler EJ. Transcriptional and epigenetic mechanisms of addiction. *Nat Rev Neurosci.* 2011; 12:623–637. [PubMed: 21989194]
141. Perrotti LI, Hadeishi Y, Ulery PG, Barrot M, Monteggia L, Duman RS, Nestler EJ. Induction of FosB in reward-related brain structures after chronic stress. *J Neurosci.* 2004; 24:10594–10602. [PubMed: 15564575]
142. Moonat S, Starkman BG, Sakharkar A, Pandey SC. Neuroscience of alcoholism: molecular and cellular mechanisms. *Cell Mol Life Sci.* 2010; 67:73–88. [PubMed: 19756388]
143. Krishnan HR, Sakharkar AJ, Teppen TL, Berkel TD, Pandey SC. The epigenetic landscape of alcoholism. *Int Rev Neurobiol.* 2014; 115:75–116. [PubMed: 25131543]